Transformation of B. burgdorferi cells

1. Vortex the cells for a few seconds (to prepare competent cells refer to protocol on competent cell preparation) and transfer 100 μL into a sterile 1.5ml microfuge tube containing the plasmid DNA (10-20 μg in a 5-10 μL volume of sterilized H2O).

2. Transfer the cell/DNA mix to a pre-chilled disposable 0.2cm-gapped cuvette. (cuvettes and cuvette holder are kept at -20ºC until use). Tap the cuvette on the side or on its bottom to get all of the cell/DNA mix settled down. Make sure to remove any condensation on the outer surface of the cuvette before electroporation.

3. Electroporate the cell/DNA mix using the following settings for the electroporator: 2.5 kV, 25 μF, and 200 Ω. A good time constant is 4.8ms or above.

3. Immediately after transformation, quickly resuspend the cells in a final volume of 5 mL BSK II medium. This is done by taking a small amount of BSK II, with a Pasteur pipette, and resuspending the electroporated cells from the cuvette and transferring them into the 5ml of BSK II. Transformed cells are generally allowed to recover overnight at 35ºC before selection is imposed. The length of the recovery period can be shortened to 8 hours, which allows transformation and plating to occur in the same day. (refer to protocol on plating for *Borrelia burgdorferi*)